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Persistence of bactericidal antibodies following infant serogroup B meningococcal immunization (4CMenB) and booster dose response at 12, 18 or 24 months of age

Matthew D Snape FRCPCH MD[1], Merryn Voysey M.Biostat[2], Adam Finn FRCP PhD[3], Gianni Bona MD[4], Susanna Esposito MD[5], Nicola Principi MD[5,] Javier Diez-Domingo MD PhD[6], Etienne Sokal MD PhD[7], Dorothee Kieninger MD[8], Roman Prymula MD PhD[9], Peter M Dull MD[10], Igor Kohl PhD[10], Michelangelo Barone MD[11], Huajun Wang MSc[12], Daniela Toneatto MD[11], Andrew J Pollard FRCPCH PhD [1] for the European Men B Vaccine Study Group.

[1] Oxford Vaccine Group, Department of Paediatrics, University of Oxford, and NIHR Oxford Biomedical Centre, Oxford University Hospital NHS Trust, Oxford, UK

[2] Nuffield Department of Primary Care Health Sciences, University of Oxford, Oxford UK

[3] Bristol Children's Vaccine Centre, University of Bristol & University Hospitals Bristol NHS Foundation Trust, Bristol, UK

[4] Azienda Ospedaliero-Universitaria, Azienda Ospedaliero-Universitaria Maggiore della Carità - Clinica Pediatrica, Novara, Italy

[5] Pediatric Highly Intensive Care Unit, Università degli Studi di Milano, Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy

[6] FISABIO-Public Health, Valencia, Spain

[7] Université Catholique de Louvain, Cliniques Saint Luc, Dept of Pediatrics, 10 av Hippocrate, B 1200 Bruxelles, Belgium

[8] Center for Clinical Studies, Department of Paediatric Immunology, University of Mainz, Mainz, Germany

[9] Faculty of Medicine in Hradec Kralove; Charles University Hradec Kralove, Czech Republic

[10] Novartis Vaccines and Diagnostics Inc. Cambridge, MA

[11] Novartis Vaccines and Diagnostics S.r.l., Siena, Italy

[12] Novartis Pharma BV, Amsterdam, The Netherlands

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SBA persistence post infant 4CMenB

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Corresponding author:

Matthew D Snape

Oxford Vaccine Group, University of Oxford Department of Paediatrics

CCVTM, Churchill Hospital, Old Rd Headington, OX3 7LE

matthew.snape@paediatrics.ox.ac.uk

Telephone/Fax+44 (0) 1865 857 420

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Abstract

Background

A serogroup B meningococcal vaccine (4CMenB) is licensed for infant use in countries including Canada, Australia and those of the European Union. Data on serum bactericidal antibody (hSBA) waning and the ideal timing of a 'toddler' booster dose are essential to optimise vaccine utilization.

Methods

An open-labeled, multicenter phase-2b follow-on European study conducted from 2009 to 2012. Participants previously receiving 4CMenB with routine vaccines at 2,4,6 or 2,3,4 months (246Con and 234Con) or at 2,4,6 months intercalated with routine vaccines (246Int) received a booster dose at 12, 18 or 24 months. 4CMenB-naïve 'Control' participants aged 12, 18 or 24 months received two doses of 4CMenB two months apart.

Results

1588 participants were recruited. At 12 months, prior to any booster doses, the proportions with hSBA titers ≥ 1:5 for strain 44/76-SL (testing vaccine component fHBP) were 73% (120/165) for the '246Con' group, 85% (125/147) for '246Int', 57% (51/90) for '234Con' and 13% (26/199) for Controls. For strain 5/99 (NadA) proportions were ≥ 96% (all 4CMenB-recipients) and 1% (Controls). For strain NZ98/254 (PorA) these were 18-35% (4CMenB-recipients) and 1% (Controls). By 24 months, 4CMenB-recipient proportions were 13%–22% (44/76-SL), 82%-94% (5/99) and 7-13% (NZ98/254) and in Controls ≤ 4%. Following a 12-month booster-dose ≥ 95% of previously immunized participants had titers ≥1:5 (all strains).

Conclusions

A 4CMenB booster-dose can overcome waning hSBA titers after early-infant immunization. Administration at 12 months could help to maintain immunity during an age of high risk, and the persistence of this response requires further study.

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Introduction

The recent licensing of a novel vaccine against serogroup B meningococcal disease (4CMenB) by the USA food and drug administration (FDA) for individuals aged between 10 and 25 years (1), by the EMA from 2 months of age in the European Union (2), and administration to over 45,000 individuals aged 2 months to 20 years in the Saguenay-Lac Saint-Jean region of Quebec (3) are important steps towards the prevention of this potentially fatal infection. It has also been recommended that the vaccine be introduced into the UK routine infant immunization schedule (4, 5). An important consideration in cost-effectiveness calculations is whether the selected schedule can maintain protection during the early years of life. This in turn is likely to be dependent on the persistence of antibodies following immunization, as an association between waning antibodies and reduced vaccine effectiveness has been seen both for conjugate vaccines against serogroup C (6) and for outer membrane vaccines against serogroup B (7-9).

In a pre-licensure study participants received 4CMenB at 2, 4, 6 months of age (alone or with routine concomitant vaccines) or at 2, 3, 4 months of age (with concomitant vaccines) (10). This study design provided the opportunity to assess the rate of bactericidal antibody waning after a range of infant 4CMenB immunization schedules, and accordingly a follow-on study was performed to assess maintenance of vaccine-induced bactericidal antibodies to 12 months of age and beyond, as well as the immunogenicity of a booster dose of 4CMenB when administered at 12, 18 or 24 months of age

Methods

Study participants and trial design

This follow-on study of a phase IIb, open-label, randomized, parallel-group controlled trial was conducted between July 2009 and January 2012 in Belgium (6 centers), UK (4 centers), Czech Republic (4 centers), Germany (24 centers), Italy (5 centers) and Spain (16 centers). Appropriate regulatory authorities in each participating country granted approval for the trial. As previously described, participants in the parent study were randomised 2:2:1:1 to receive: 4CMenB concomitantly with routine immunization at 2, 4, and 6 months of age (246Con), 4CMenB at 2, 4, 6 months of age, intercalated to routine vaccines at 3, 5, 7 months of age (246Int), 4CMenB with routine vaccine at 2, 3, 4 months (234Con) or routine vaccines alone (Control)(10). At the last visit in this study participants' parents were invited to enrol their child into the follow-on study. Following parental informed consent, enrolled 'follow-on' participants who previously received 4CMenB were randomised 1:1:1 to receive a booster dose of 4CMenB at 12, 18 or 24 months of age (see Figure1) according to a centrally held randomization list (Novartis) using fixed block size of 3. Randomization in the follow-on study occurred prior to enrollment based on the participants completing the parent study, but follow-on randomization group was to have been concealed from parents up until the time of enrollment. Those who previously received routine vaccines alone received 4CMenB at 12 and 14 months of age (Control 12, 14). Additionally, two groups of 50 'Control' participants who were 4CMenB naïve were to be recruited to receive 4CMenB at 18 and 20 (Control 18, 20), or 24 and 26 months of age (Control 24, 26). Blood tests were taken immediately before the first dose of 4CMenB and one month after each dose.

Exclusion criteria were any history of meningococcal infection or close contact with someone experiencing meningococcal disease, prior receipt of meningococcal B (control participants

only) vaccines, acute or chronic illness, known reactions to vaccine components, known or suspected immune disease or impairment including the administration of steroids, receipt of antibiotics 6 days prior to enrollment, receipt of blood products or planned receipt of non-study vaccines (supplementary Table 1)

In the UK control participants were recruited by information letters sent via National Health Service child health computer databases while those in other study centers were recruited through paediatric hospitals or private practices.

Intervention

4CMenB consists of 50µg each of Neisserial adhesin A (NadA), and fusion proteins containing Neisseria Heparin Binding Antigen (NHBA) and Factor H Binding Protein (fHbp), 25 µg detoxified OMV from *N. meningitidis* strain NZ98/254, 1.5 mg aluminium hydroxide and 10 mM histidine in 0.5 ml water for injection. All vaccines were administered by intramuscular injection in the antero-lateral thigh. In order to maintain participant's immunizations in line with their country's immunizations programme, routine immunizations were offered as per supplementary Table 2, but did not form part of the study evaluation. These vaccines were not administered within 30 days of the study vaccines.

Functional antibody

Serum bactericidal activity using human complement (hSBA) was assessed at the Novartis Vaccines Serology Laboratory, Marburg, Germany against three reference strains chosen to determine the immunogenicity of individual vaccine components—strain 44/76-SL for fHbp, strain 5/99 for NadA and strain NZ98/254 for OMV. A strain not included in the original study (M10713) was used to assess the immunogenicity of NHBA as previously described(11). hSBA was expressed as interpolated titers according to reciprocal serum

dilutions yielding 50% or greater killing of the target strain after 60 minutes of incubation compared with growth at time 0. An interpolated titer ≥ 1:5 represented 95% confidence that participants achieving this titer had a protective hSBA (≥1:4).(12)

Safety

Parents recorded local injection site (pain, erythema, swelling and induration) and systemic reactions (i.e. fever [axillary temperature ≥ 38°C], change in eating habits, sleepiness, unusual crying, vomiting, diarrhea, irritability, rash) for seven days after each vaccination. Injection site pain was classified by the parents as mild (minor reaction to touch), moderate (cried or protested when touched), or severe (pain on limb movement). Erythema, induration and swelling were summarized by maximal severity (1-25 mm, 25-50 mm and >50 mm). Adverse event recording was enhanced by telephone contact in the week after study vaccination. Safety follow-ups were completed 6 months after the last dose of 4CMenB. All serious adverse events (SAEs) reported during the study were recorded. Determination of the relationship between adverse events and the study vaccine was made by the study site's investigator judgment based on temporal relationship and biological plausibility criteria.

Statistics

The pre-specified population for the immunogenicity analysis was the per-protocol (PP) population, analysed as per-immunization course received and including all those receiving all the relevant vaccine doses correctly, provided evaluable serum samples at the relevant timepoints and had no protocol violations having a significant impact on immunogenicity analysis.

The proportions of participants with hSBA≥ 1:5 for the reference strains were calculated, as were the 2-sided 98.3% Clopper Pearson confidence intervals (CI). In addition, hSBA titers

were log transformed and their geometric mean titers (GMTs) and 2-sided 95% CI calculated, as were the between-group ratios of GMTs and 2-sided 95% CIs (using two-way analysis of variance with a factor for vaccine group and country).

The primary outcome was the proportion of participants in the 246Con group with hSBA titers ≥ 1:5 for strains 44/76-SL, NZ98/254 and 5/99 following a booster dose of 4CMenB at 12, 18 or 24 months of age. A sufficient immune response was pre-specified as the lower limit of the 98.3% confidence interval for this proportion being above 75% for at least one of the booster dose timings. Based on previous immunogenicity studies, indicating a likely 'true' rate of 90% for participants having hSBA ≥ 1:5 (13), performing 5000 simulations suggested a sample size of 180 participants would provide 99% power to demonstrate this.

The main secondary outcomes were the proportions with hSBA titers ≥1:5 and hSBA GMTs before the booster dose in the above participants, and before and after the booster dose for 246Int and 234Con participants. The induction of immunological memory was assessed by comparison of the response to a single dose of 4CMenB at 12, 18 or 24 months in participants previously immunized in infancy and those who were MenB vaccine naïve. The immunogenicity of 2 doses of 4CMenB given at 12 and 14, 18 and 20 and 24 and 26 months was also assessed. The above measures of hSBA activity against strain M10713 were determined as a post-hoc analysis.

Individuals who received at least one dose of vaccine and provided post-baseline safety data were included in the safety analyses, for which results were reported descriptively with no formal statistical analyses. Safety objectives were the tolerability of a booster dose of 4CMenB at 12, 18 or 24 months of age, and of a 2 dose schedule commencing at these ages in MenB vaccine naïve children.

Statistical analysis was performed using SAS version 9.1. (SAS Institute, Cary, NC).

Results

1799 participants completed the parent study, of whom 1481were enrolled into this study and randomised to a booster dose at 12, 18 or 24 months of age. In addition, 51 new participants were recruited at 18 months, and 56 at 24 months (Figure 1). Of these 1588 participants, 826 (52%) were male and 94% were Caucasian.

1495 (94%) participants completed the study, the withdrawals primarily being due to withdrawal of consent (41), loss to follow up (26), protocol deviations (14) and inappropriate enrollment (7). There was one death in a child following a car accident, prior to receipt of study vaccine. The PP population for immunogenicity included 1221 (76%) of enrolled participants, and the safety analysis population included 1519 (96%).

Immunogenicity

Primary outcome

The proportion of 246Con participants with hSBA ≥ 1:5 following a booster dose of 4CMenB at 12 months of age was 97% (93 – 99) for strain 44/76-SL, 100% (97 – 100) for strain 5/99 and 95% (89 – 98) for strain NZ98/254 (Table 1). As the lower 98.3% confidence limit for these proportions was above 75% for all three strains, the primary objective for the study was met. hSBA titres for this group following a booster dose at 18 and 24 months are also shown in Table 1 (a).

Secondary objectives

The proportions with hSBA ≥ 1:5 following the booster dose of 4CMenB administered at 12, 18 and 24 months to participants in groups 246Int and 234Con were similar to those in the

246Con group outlined above, being 98% or greater for strains 44/76-SL and 5/99, and 80 to 97% for strain NZ98/254 (Table 1 (a)).

Prior to administration of the booster doses at 12, 18 and 24 months of age progressive waning of these proportions was observed for all groups for strains 44/76-SL and NZ98/254, with waning particularly rapid for the latter strain (no greater than 13% by 18 months of age). In 4CMenB vaccine naïve control participants, prior to immunization the proportion of participants with hSBA ≥ 1:5 were no higher than 13% for strains 44/76-SL, NZ98/254 and 5/99.

For the control participants, a two-dose immunization schedule resulted in at least 91% of recipients having hSBA ≥ 1:5 for strains 44/76-SL, 5/99 and NZ98/254.

As a post-hoc analysis, strain M10713 hSBA titers for a subset of participants in the 234Con group and the Control groups were determined. Pre-booster dose, the proportions of these participants with hSBA ≥ 1:5 were similar regardless of whether they had been primed with 4CMenB or not. (Table 1 (a))

hSBA GMTs are demonstrated in Figure 2 and Table 1 (b). For all strains, a greater rise in hSBA GMTs was seen following a single dose of 4CMenB in participants previously primed with this vaccine than in control recipients.

All control participants receiving two doses of 4CMenB achieved hSBA ≥ 1:5 for strains 44/76-SL and 5/99; for NZ98/254 this proportion ranged from 91% to 99%, and for M10713 was 86% (Control 18, 20) and 81% (Control 24, 26).

Rates of fever following a single dose of 4CMenB ranged from 20% to 45% and were similar in children receiving 4CMenB for the first or fourth time (Figure 3). A trend to increasing reported rates of severe tenderness at the injection site with increasing age was also apparent

(Figure 3); again, this was not influenced by the number of previous doses of 4CMenB received.

During the study 70 serious adverse events affecting 58 participants were reported (including the death mentioned above). Three of these were classified by investigators as possibly being related to the study vaccine. One child enrolled into the Control 12, 14 month group, was diagnosed with autism at the age of 3 years, having initially presented with speech delay and learning difficulties at age 18 months. This child was immunised with his routine vaccines (as per UK schedule) at 12 and 14 months of age, and with 4CMenB at 15 and 17 months of age. The investigator considered it was not possible to exclude a relationship with the study vaccine given the temporal association. This child's previous medical history was unremarkable, and speech and hearing assessments were normal. At 3 ½ years of age the child had persistent difficulties with language development and social interaction. A second child, in the 246Con-24 group, developed idiopathic epilepsy, with the first convulsion at age 2 years and 3 months, 106 days after the booster dose of vaccine at 24 months of age. EEG and MRI tests were normal. By the age of 3 ½ years the child had experienced six seizures (both febrile and afebrile), but none within the last 3 months. There was no developmental delay and the child was not receiving anticonvulsant therapy. A third child (Control 12, 14) developed two febrile convulsions on the day of the 12 month immunization, and was also diagnosed with atypical pneumonia at the time of presentation.

Discussion

This study of over 1500 participants provides novel data on the persistence through the second year of life of bactericidal antibody induced by various infant 4CMenB immunization regimens and on the immunogenicity and reactogenicity profile of 4CMenB given as two doses in the second year of life. These data, combined with local age-specific epidemiology,

are vital for the design of the optimal strategies for implementation of 4CMenB immunization programmes.

The UK Joint Committee on Vaccines and Immunization (JCVI) recently recommended that 4CMenB be introduced in a 2, 4, 12 month immunization schedule into the routine infant immunization program (4, 5). Specific studies of the immunogenicity of this '2 +1' schedule are currently underway(14). However, the European Medicines Agency marketing authorisation for the vaccine is for a 3-dose priming schedule with a booster in the second year of life (3+1), a schedule which was informed by the data presented in this study(2) and which may be selected by some national authorities.

Importantly, in the current study the proportions of participants with SBA titers $\geq$ 1:5 prior to boosting in the second year of life was not influenced by whether the last priming vaccine was given at 4 or 6 months of age. For both schedules the response to a single booster dose of 4CMenB was also similar and this, combined with the relatively lower hSBA GMTs following the first dose of 4CMenB in control participants, suggests both schedules effectively induced immunological memory.

These data provide reassurance that 4CMenB could be incorporated into either a 2, 4, 6 month schedule (as used in North America, Latin America and many European countries) or into an accelerated 2, 3, 4 dose schedule without impacting on the persistence of immune protection through the first year of life or following the booster dose.

The study also informs the optimal timing of the booster dose. The ongoing waning of antibodies observed from 1 to 2 years of age in the second year of life is of some concern given the incidence of serogroup B meningococcal disease at this age is second only to infancy in Europe(15) and suggests that where this is the case booster dose should not be delayed beyond 12 month. Whether or not this disadvantage could be offset by greater

persistence of antibodies through pre-school years following a delayed dose is not currently known, and is being evaluated in a further persistence study(16).

The licensed schedule for 4CMenB includes two doses given two months apart for children aged 12 months to 10 years, with a booster dose given 1 to 2 years later for children initially immunized at 12 to 23 months (17). These data demonstrate that, although 88% or more of children developed SBA titres ≥ 1:5 for strains 44/76 and 5/99 after a single dose, a two dose priming schedule is required to be immunogenic for all 4 strains, supporting the licensed schedule for his age group. There was little apparent difference in immunogenicity across this age band.

There was also little difference in the reactogenicity profile between controls immunized at 12, 18 or 24 months, with the exception of a trend to higher rates of reported severe tenderness in older children. This trend was also noted with increasing age of the booster dose. It is possible that this reflects a trend to greater local reactogenicity with greater age, however an alternative possibility is that severe tenderness (i.e. tenderness on limb movement) might be more readily reported as the developing child becomes increasingly verbal. The rates of fever following 4CMenB given in the second year of life were similar to those reported when this vaccine was given without concomitant vaccines in infancy (10), however they are higher than those reported following glyco-conjugate vaccines such as MenC given to one to two year olds (18-20). This tendency for the vaccine to be relatively reactogenic compared to other routinely used childhood immunisations has been described in previous clinical trials, and in the UK parents are advised to routinely administer paracetamol prophylactically when their child receives their 2 and 4 month 4CMenB immunisations(21). No such recommendation is given for the 12 month booster immunisation, although this was accompanied by fever rates of 30% to 45% in our study.

The bactericidal titers against the M10713 strain, included as a post-hoc assessment of the immunogenicity of the NHBA component, warrant specific consideration. Only 36% of participants developed bactericidal antibodies against this strain following a 2, 3, 4 month schedule, and hSBA titres were similar to those of controls following priming and prior to boosting. Direct comparisons of immunogenicity against M10713 for 234 and 246 schedules were not possible within this study, however, for a previously reported study over 80% of participants receiving a 2, 4, 6 schedule had hSBA $\geq$ 1:5 for this strain, which was maintained at 60% prior to boosting at 12 months(22). Within the limits of cross-study comparisons this does suggest that, for this antigen at least, a 2, 4, 6 month schedule may be preferable. It is worth noting that, as for the other strains, a greater rise in M10713 specific bactericidal antibodies was seen among primed infants than in those receiving this for the first time, again suggestive of immunological memory.

The data obtained in this study on antibody persistence after primary immunization in the absence of a booster dose, complement those obtained in other studies after a 4CMenB toddler booster dose (11, 23), a further booster dose at 3 ½ years of age (24), primary immunization at 3 ½ years of age (24) and primary immunization in adolescence (25). As with meningococcal conjugate vaccines (26), antibody persistence appears to be enhanced with increasing age at primary immunization.

Another feature is the variable rate of waning between different strains, with bactericidal titers against strain 5/99 (NadA) being maintained in the majority of vaccine recipients while those against NZ98/254 (PorA) waned more rapidly, and an intermediate rate of waning observed for strains 44/76 (fHBP) and M10713 (NHBA). This pattern of SBA waning has been highly consistent across multiple studies (11, 23, 24, 27) . In the absence of any data on whether relatively persistent SBA titres are reflected in more prolonged immunity against strains bearing the relevant antigens, is unclear whether this is likely to be clinically relevant

or merely reflects different susceptibilities of the strains to killing in the serum bactericidal antibody assay.

This differential rate of waning has made predicting persistence of the immune protection provided by immunization with 4CMenB particularly difficult. The interim statement by the JCVI published in July 2013 suggested that infant immunization with this vaccine would directly prevent approximately 25% of the lifetime risk of invasive MenB disease in the UK (28). Should the vaccine be introduced into routine infant immunization, extensive post-implementation surveillance is already planned to monitor the accuracy of such predictions (29), and whether strains bearing antigens targeted by 'waning' antibodies will be over-represented as a cause of secondary vaccine failures.

A limitation of this study was the lower numbers of participants at the 18 and 24 month time points. This in part reflected an increasing number of withdrawals with advancing age of booster immunization, reflecting greater interval between enrollment and immunization, but also appears to reflect a discrepancy in numbers recruited to different randomised groups at the conclusion of the parent study. This may in turn reflect a problem with concealment of randomisation group from the parents prior to enrollment. Of note is that although the routine vaccines administered during the study did differ between countries, these vaccines were not administered within 30 days of the study vaccine. In the parent study we demonstrated that routine glyco-conjugate and diphtheria, tetanus, pertussis and polio vaccines had minimal impact on 4CMenB immunogenicity, even when administered concomitantly(10). It is therefore highly unlikely these variable routine immunization schedules would have influenced the 4CMenB immunogenicity profile in this study.

Nevertheless, this study makes an important contribution to the expanding body of knowledge regarding 4CMenB. The recent 4CMenB mass immunization campaigns at

Princeton University (30), University of California, Santa Barbara (31) and Saguenay-Lac Saint Jean, Quebe(3), highlight the importance of a vaccine being available against this rapidly fatal infection. Information on duration of immunity is essential to plan schedules and provide understanding of how to best use this vaccine. Our data support the use of a booster dose early in the second year of life following infant immunization, and the need for further studies to look at ongoing antibody persistence beyond the time points measured in this study. Nonetheless, as with other vaccines licensed based on immunologic surrogate measures, a firm understanding of the duration of protection will only be established after broad vaccine implementation with robust disease surveillance and vaccine coverage assessments.

Footnotes

Declaration of Interests: M D Snape, A Finn, S Esposito, N Principi, D Kieninger, J Diez-Domingo, E Sokal, R Prymula act as investigators for clinical studies from both non-commercial funding bodies and commercial sponsors (i.e. some or all of Novartis Vaccines, GlaxoSmithKline, Sanofi-Aventis, Sanofi-Pasteur MSD, MedImmune and Pfizer Vaccines) conducted on behalf of their institutions as listed in the affiliations. MD Snape and A Finn participate in advisory boards and speaking engagements for vaccine manufacturers; all payments received are paid to their respective institutions. R Prymula, J Diez-Domingo, S Esposito and N Principi also undertake consultancy and advisory work and receive speaking honoraria, travel and accommodation reimbursements for several commercial sponsors. The NIHR Oxford Biomedical Research Centre provides salary support for M D Snape, who is a Jenner Investigator. A J Pollard is a Jenner Investigator and James Martin Senior Fellow. A J Pollard and A Finn do not receive any personal renumeration from vaccine manufacturers. AJ Pollard is chair of the UK Department of Health's (DH) Joint Committee on Vaccination and Immunization (JCVI); the views presented in this manuscript do not necessarily represent the views of DH or JCVI. P Dull was formerly an employee of Novartis Vaccines and Diagnostics, D Toneatto, H Wang, I Kohl and M Barone are employees of Novartis Vaccines and Diagnostics.

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her. M Voysey is independent of the Sponsor and is employed by the University of Oxford and was not compensated by Novartis.

Statistical and Data Management Center: Statistical evaluation of the results was performed by Novartis Vaccines and confirmed by an independent statistician at Oxford University working with the Nuffield Department of Primary Care Health Sciences.

Presentations at Meetings

The data presented in this manuscript have been presented at the European Society of Paediatric Infectious Diseases Meeting in Milan, Italy May 2013.

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Author Contributions:

Study concept and design: A J Pollard, M D Snape, P Dull, D Toneatto, S Esposito, E Ypma, N Principi, R Prymula,

Acquisition of data: E Sokal, J Diez-Domingo, MD Snape, S Esposito, A J Pollard, A Finn, G Bona, N Principi, R Prymula, B Becker, D Kieninger; I Kohl, Michelangelo Barone

Analysis and interpretation of data: M D Snape, A J Pollard, D Toneatto, P Dull, S Esposito, E Ypma, G Bona, M Voysey, N Principi, H Wang,

Merryn Voysey had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Drafting of the manuscript: MD Snape,

Critical revision of the manuscript for important intellectual content: A J Pollard, D Toneatto, J Diez-Domingo, E Sokal, M D Snape, S Esposito, G Bona, M Voysey, N Principi, D Kieninger, R Prymula, B Becker A Finn, B Becker, P M Dull, I Kohl , M Barone, H Wang

Statistical analysis: Huajun Wang, Merryn Voysey

Administrative, technical, or material support: E Sokal

Study supervision: A J Pollard, P Dull, D Toneatto, M D Snape, S Esposito, G Bona, N Principi, R Prymula

The European Men B Vaccine Study Group:

UK: St George's University of London, Division of Child Health: Paul T Heath MD, Clarissa Oeser MD; MCRN South West Royal Cornwall Hospitals NHS Trust: Andrew Collinson MD, Jasmine Heslop RN; University of Oxford, Department of Paediatrics, Oxford Vaccine Group: Tessa John RN, Sarah Kelly RN; Italy: University of Florence, Department of Sciences for Woman and Child's Health, Anna Meyer Children's University Hospital: Maurizio De Martino MD, Luisa Galli MD; Chiara Azzari MD, Leila Bianchi MD; University of Padua, Department of Pediatrics: Carlo Giaquinto MD, Susanna Masiero MD; Milano: Pediatria dell'Ospedale Sacco di Milano: Gianvincenzo Zuccotti MD; MA and Siena: Novartis Vaccines: Benedetta Ghezzi, Igor Kohl, Frank Baehner, Carina Nasemann, Simone Inzillo, Riccardo Belli, Lucie Hlavata; Azienda Ospedaliero-Universitaria, Azienda Ospedaliero-Universitaria Maggiore della Carità - Clinica Pediatrica, Novara, Italy; Gianni Bona MD, Nicolino Grasso MD; Spain: Castellón: Centro de Salud San Lorenzo: Miguel A Cabañero MD; Centro de Salud Almassora: Eva Suárez MD; Centro de Salud Vall D'Uixo: Susana Peñarroja MD; Valencia: Centro de Salud Raval Sagunto: Vicente Antón MD, PhD; Centro de Salud República Argentina: Manolo Martínez MD; Centro de Salud Nazaret: María D Garcés MD; Centro de Salud Malvarrosa: Angels Jubert MD; Centro de Salud Serreria I: Maite Asensi MD; Centro de Salud Serreria II: Ignacio Sorribes MD; Centro de Salud de La Eliana: Isabel Úbeda MD, PhD; Centro de Salud Paiporta: Victoria Planelles MD; Centro de Salud Plaza de Segovia: Jose Villarroya MD; Centro de Salud Catarroja: Miguel Tortajada-

Gribes MD, PhD; Santiago de Compostela: Hospital Clínico Universitario de Santiago de Compostela: Federico Martinón-Torres MD PHD, Lorenzo Redondo Collazo MD; Oviedo: Hospital Universitario Central de Asturias, Department of Paediatrics: Julián Rodriguez S. MD PHD; Belgium: Antwerp: Campus Sint-Vincentius: André Vertruyen MD, GZA; University Hospital Antwerp: Jose Ramet MD; Namur: Centre hospitalier regional de Namur: Marc Verghote MD; Hasselet: Virga Jesse Ziekenhuis: Marc Raes MD; Brussels: Clinic of Pediatric Respiratory Diseases, Infectious Diseases and Travel, UZB - Universitair Ziekenhuis Brussel, Department of Pediatrics: Anne Malfroot MD; Germany: NETSTAP e.V., Bochum: Ulrich Behre MD, Martin Kimmig MD, Roland Knecht MD, Dieter Schlegel MD, Siegfried Simmet MD, Michael Steiner MD, Bernhard Sandner MD, Martina Weh MD, Eckhard Ziegler-Kirbach MD, Friedrich Kaiser MD, Gerhard Bleckmann MD,Thomas Adelt MD, Ralph Köllges MD, Heidemarie Pankow-Culot MD, Peter Soemantri MD, Philip Fellner von Feldegg MD, Dietmar Hauptmann MD, Christian Weißhaar MD, Roland Achtzehn MD, Christian Horn MD, Christoph Schäfer MD, Brigitte Wilmsmeyer MD, Eivy Franke-Beckmann MD; Czech Republic: Hradec Kralove: University of Defense, Faculty of Military Health Sciences, Vaccination Center: Roman Chlíbek MD PHD; Pardubice: Children´s department Pardubice: Vladimír Němec MD; Cerveny Kostelec: Detske stredisko: Luděk Týce MD; Jindrichuv Hradec: Samostatna ordinace praktickeho lekare pro deti a dorost: Daniel Dražan MD;

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Figure 1: Study design and participant numbers

Abbreviations: rand, randomized; imm, immunized; comp, completed.

Routine vaccines in the parent study were a combination diphtheria, tetanus toxoid, acellular pertussis, *Haemophilus inflluenza* type b, Hepatitis B and inactivated polio vaccine (DTapHib-HepB-IPV) and 7-valent pneumococcal vaccine.

Figure 2:

Serum bactericidal activity geometric mean titers by strain and age at booster dose

Figure 3:

Rates of systemic (figure 3a) and local (figure 3b) reactions following immunization at ages shown.